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SOME APPLE DISEASES

THE VOLUTELLA ROT

F. L. Stevens + J. G. Hall

N. C. COLLEGE OF AGRICULTURE AND MECHANIC ARTS.

THE NORTH CAROLINA

AGRICULTURAL EXPERIMENT STATION

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PRACTICAL SUMMARY.

Several apple diseases, part of them not heretofore recognized, are described in this bulletin. They consist of two fruit rots, two twig blights and a bark disease which is called the scurf.

These diseases are all caused by fungi and are therefore probably amenable to treatment with the ordinary sprays. The existence of some of these fruit rotting fungi in winter upon twigs indicates the necessity of carefully pruning out all dead twigs to prevent infection of the fruit, and of other twigs, in following seasons.

Spraying, rendered necessary by old and too well known apple diseases, is made doubly important by the discovery of these new enemies to apple culture.

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THE VOLUTELLA ROT.[1]

BY F. L. STEVENS AND J. G. HALL.

A black rot of apples, closely imitating in appearance that caused by Sphaeropsis, but differing from the Sphaeropsis rot in several details, has been observed frequently in various sections of this State, on native apples and on apples shipped into the State from a distance.

In general appearance the disease consists of a rotten black spot, frontispiece, the spot increasing in size until it eventually encompasses the whole fruit. The central and older portions of the decayed region are of an intense coal black color. The younger region of the spot, its outer border, a zone about five-eighths of an inch wide, is brownish.

Close inspection reveals the presence of slightly elevated pimple-like places in the cuticle. These are found to within about one-fourth of an inch of the edge of the spot, and become larger and more pronounced as the center of the spot is approached, indeed, the black color of the spot seems to be due to the thick setting of these black pimples all over its surface. In many instances, unless the spot be quite old, no other development is seen, and the disease might readily

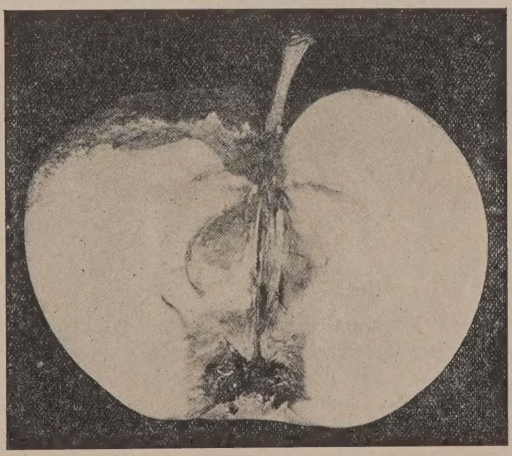

FIG. 2. The Volutella Rot in Section.

be considered to be the ordinary black rot, caused by Sphaeropsis, and doubtless often passes for it. In older spots, however, the pimples

1 Published in part in Journal of Mycology vol. 13, p. 94, May, 1907.

are seen to have broken through the cuticle of the apple, and each pimple appears as a small wart-like excrescence, and a good lens shows that it is thickly beset with stiff black hairs. These hairs constitute the distinctive character of this disease, and serve to separate it with ease and certainty from the Sphaeropsis rot, provided the rot is developed far enough to exhibit this character.

On slicing the apple open it is seen that the zone most recently invaded is brownish, while all of the older portion is black. (Fig. 2.)

While the decayed portions are softer than the healthy, this is in no sense a wet rot, the softness being due to a spongy dryness rather than to any watery breaking down of tissue. Upon microscopic examination the cells of the old dark, diseased portion of the apple are found to be filled with a tangled mass of black or dark brown fungous threads (Fig. 3), which are richly septate and much branched. In

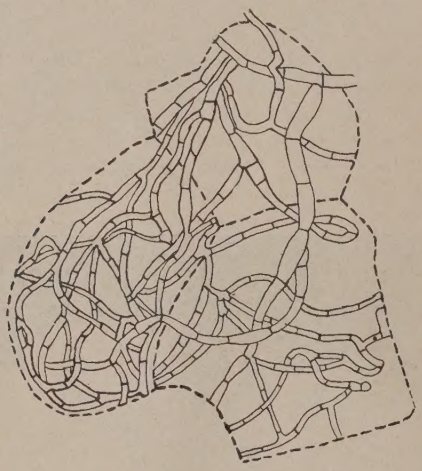

FIG. 3. Cell Invaded by Mycelium.

thickness they vary from 5 to 7 Mu[1] in the older portions. In the newly invaded cells the mycelium is usually only about 2 Mu thick and is colorless.

As the spot ages the mycelium develops more abundantly in a few layers of cells next to the cuticle, particularly immediately under the cuticle. From this sub-cuticular layer of tangled mycelium there develops a cushion of hyphae, which are arranged parallel and stand upright, perpendicular to the substratum. These hyphae are quite uniform in thickness and regular in arrangement, composed of short

[1] 1 Mu equals 1-25,000 of an inch.

rectangular cells. (Fig. 4.) These upright hyphae increase in length, rupture the cuticle and develop the tubercular mass, (Fig.

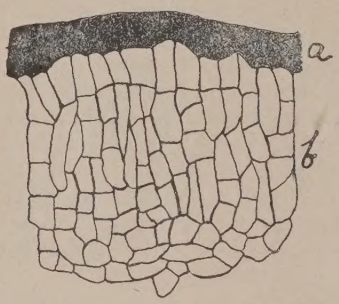

FIG. 4. Palisade Arrangement of Hyphae.

5), "sporodochium," characteristic of the fungous order Tuberculariales. The tubercle attains a height of 100 to 125 Mu and a diameter nearly twice as great. About midway from base to top of the sporo-

FIG 5. Sporodochia in Section.

dochium the hyphae become narrower and the setae have their insertion (Fig. 6). The setae develop directly from hyphae which stand amid sporogenous hyphae and are indistinguishable from them, their only point of difference being that the setigerous hyphae broaden out at the end and develop into typical setae while the sporogenous proceed to spore development. Each setum is produced from the tip of a single hypha. The setae are from 100 to 400 Mu long, tapering gradually from base to tip. At base they are 5 to 8 Mu broad. They have from 2 to 5 septa (Figs. 6 and 7).

At the upper end of the erect fuscous or black hyphae, which constitute the sporodochium, are simple, slender hyaline stalks, "conidiophores," from 25 to 35 Mu x 3 Mu long, which bear the spores (Fig. 8). The conidiophores together constitute the outermost layer of the sporodochium.

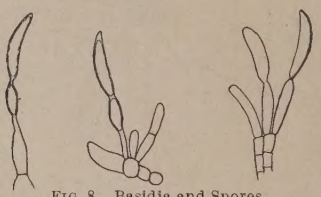

FIG. 8. Basidia and Spores.

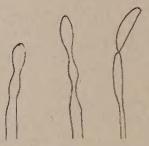

FIG. 9.
Basidia and Spores

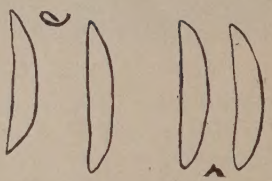

FIG. 10. Spores.

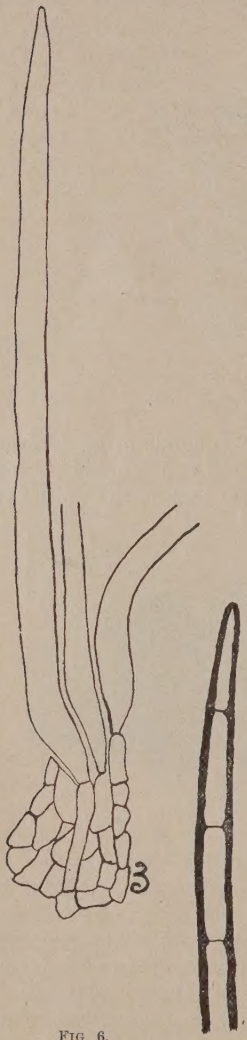

FIG. 6.
Showing Origin of Seta.

FIG. 7. Seta and Septae.

The spores are produced acrogenously, being cut off from tips of the conidiophore by a constriction of the fertile hyphae (Fig. 9).

The spores are oblong-fusoid to falcate-fusoid, with acute ends (Fig. 10), hyaline or very slightly olivaceous, continuous, though with the low power of the microscope they often appear uniseptate,

due to peculiarities of the protoplasm in this region. They measure from 17 to 23 Mu long by 2.5 to 3.5 Mu broad.

Placed in apple agar in hanging drop cultures they germinate in about three hours. At first by a single hyphal thread near one end of the spore (Fig. 11), but later other points of germination can be seen. When germination begins the spores become very coarsely granular. The granules then become fewer in number and larger as germination proceeds. Later, by the migration of the granules into the young hyphae, clear spaces appear. At first there are no septae in the hyphae, but after about 25 to 30 hours a swelling is usually seen at the end of the hypha (Fig. 11), which soon becomes cut off by a wall. This cell rapidly assumes a dark color and has the appearance of the secondary spores, so characteristic a feature in the germination of the anthracnoses. As with the anthracnoses this body may remain without further development or it may germinate immediately (Fig. 12). The growth of the hypha is slow, as shown by Fig. 11. These drawings were made from the

FIG. 11. Germinating Spores.

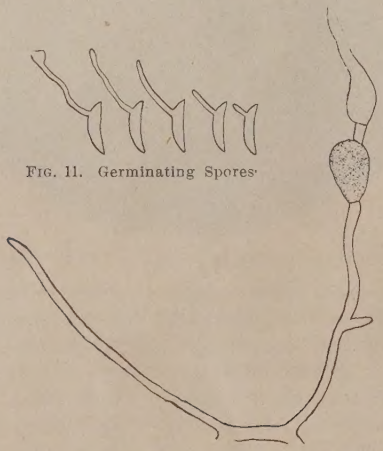

FIG. 12. Germinating Spore and Secondary Spores.

same spore, growing in a hanging drop culture, at periods of one hour.

Transfers into solidified pea agar and apple agar plates were made under aseptic conditions from the regions immediately beneath the skin in a diseased spot; also from the diseased tissue near the boundary between diseased and healthy portions, and in every instance pure cultures of the fungus were obtained. By plating from water containing many of the spores pure cultures were also obtained.

When grown in media containing starch or glucose the mycelium of the fungus is densely black. In the absence of these nutrients it

is colorless. In none of the artificial cultures were spores formed. On sterilized apple twigs, however, spores were produced in abundance.

FIG. 13. Colony on Meduim Rich in Carbohydrate

The effect of carbohydrate upon the color of mycelium is most strikingly brought out in petri dish cultures, as is shown in the accompanying illustration, representing colonies grown upon pea agar and colonies grown upon apple agar. Upon apple agar the colonies are densely black throughout (Fig. 13). Upon pea agar they are colorless, the only blackening being located in scattered spots (Fig. 14).

Inoculations were made from mycelium grown in pure culture by pricking the skin of the apple and laying on, or inserting, a bit of the mycelium. Within nineteen days of the inoculation the spot had attained a diameter of seven-eighths of an inch. The accompanying illustration (Fig. 15) shows the rapidity of development in one instance at room temperature, 101 hours intervening while the rot progressed from the inner to the outer inked circle.

Many attempts were made to secure inoculation through unbroken cuticle, but in no instance with success. It is evident that a bruise or break of some kind is necessary. This evidence correlates well

with the fact that the disease, as is true of so many fruit diseases, is much more common at the blossom end or the stem end than at other points. The rot occurs at either end of the fruit more often than elsewhere, because it is at these places that the cuticle is most liable to injury.

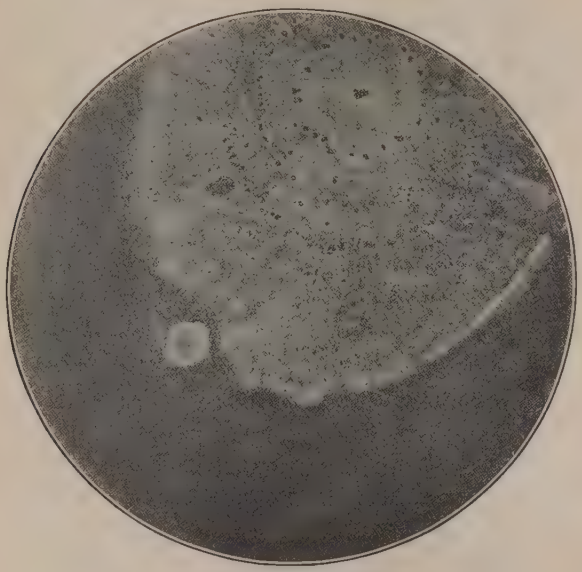

FIG. 14. Colony on Medium Poor in Carbohydrate.

The tubercular development places the fungous unquestionably among the Tuberculariales. Further, the road is not so clear. Judging strictly from the color of the mycelium it must go to the Dematiae, but a glance over its possible kin there reveals none which are of certain affinity. Admitting that it is possibly of the Mucedineae, it clearly must belong to the genus Volutella. This genus is made up mainly of light colored fungi, though several of them are very dark, notably V *Acalyphae, oxyspora, Violae, Citrulli* and *Allii*. From a structural viewpoint this fungus seems much closer to Volutella than to any of the Dematiae, and in view of the purely physiological nature of the color character, as shown by our cultures, it being dependent upon the presence of carbohydrates (which are present in abundance in the natural medium, the apple), we believe we are doing no violence to facts in placing it in the genus Volutella.

As a Volutella it stands closest to the species named above, yet is dstinct enough from them to be regarded as a separate species.

It differs from *Oxyspora* in larger sporodochia 150-400 Mu and longer setae, shorter spores, longer basidia; from *Cittrulli* in shape of spores; from *Allii,* sporodochia larger, not convex or elongate, setae

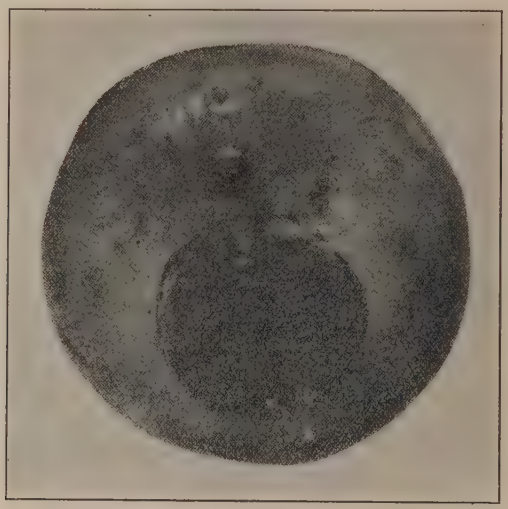

FIG. 15. Showing Rapidity of Development.

longer; from *Violae*[1]; in character of sporodochium; from *Acalyphae* in size of spores.

VOLUTELLA FRUCTI. STEVENS AND HALL.[2]

Sporodochia numerous in concentric circles early sub-cuticular, then erumpent, elevated from 200 to 250 Mu black. 150-400 Mu in diameter mycelium, black in presence of carbohydrates, otherwise hyaline.

Setae; distributed throughout sporodochium, black 0-3 septate, acute, smooth, 100-400 Mu long, 5 to 8 Mu thick.

Sporophores; elongate, hyaline, simple, 25-35 x 3 Mu.

Conidia; hyaline or very slightly olivaceous, continuous, smooth, oblong-fusoid to falcate-fusoid, 17-23 x 2.5-3.5 Mu.

Habitat; Fruit of *Pyrus Malorum,* North Carolina, Type N. C. Agricultural Experiment Station, No. 780.

TREATMENT.

It is probable that the sprayings usually recommended for scab and black rot will be effective in controlling the Volutella Rot.

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[1] See Bot. Gaz., XXVI, Pl. XIV, 85.

[2] Originally described in Journal of Mycology 13, p. 91, May, 1907.

CONIOTHYRIUM AS A FRUIT ROT.

This fungus has frequently been seen causing rotten spots in apples in this State. It was first made the object of study in this laboratory in November, 1906. The rotten spot is circular in uniform tissue, brownish, about the color of the common Penicillium rot and, due to loss from evaporation, the tissue shrinks as the spot ages, leaving the surface much wrinkled (Fig. 1). Distributed somewhat sparsely

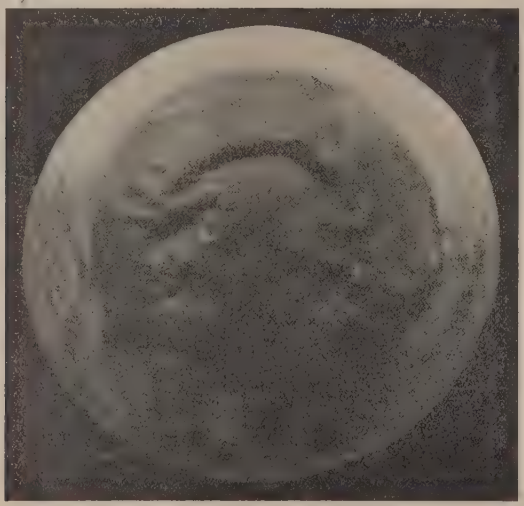

FIG. 1.—Coniothyrium Rot.

and irregularly over the spot are the fruiting cavities of the fungus "pycnidia." In rotten spots produced by inoculation the pycnidia frequently assume a somewhat concentric arrangement. Proceeding from the innermost pycnidia toward the edge of the spot the pycnidia become smaller, none at all being found nearer than about 5 mm. to the edge of the diseased tissue.

Upon sectioning through a diseased spot the flesh of the fruit is

seen to be rotten, brownish and soft (Fig. 2), not black or dry, as is the case with the Volutella Rot.

Microscopic examination shows the diseased tissue to be thoroughly

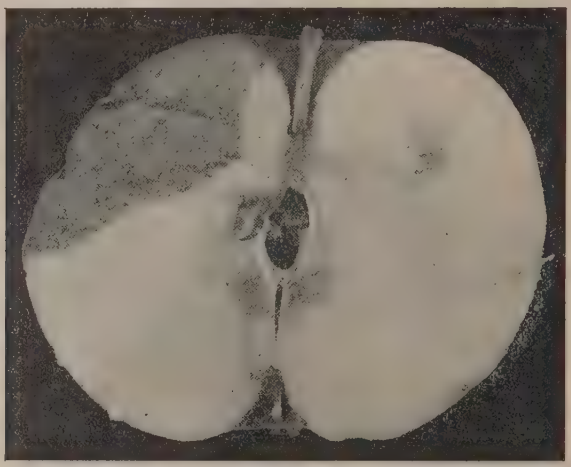

FIG. 2.—Coniothyrium Rot in Section.

invaded with a thin branching septate mycelium which, when young, is hyaline, when old fuscous or straw colored.

The fruiting bodies lie immediately under the cuticle and consist of spherical cavities about 200 Mu in diameter, walled in by a weft of mycelial threads. When mature this pycnidium bears very numerous small black or brown spores 3-5.8 x 2-2.5 Mu (Fig. 3), approximately 250,000 spores being borne in a single pycnidium. The spores are borne on stalks standing perpendicular to the wall of the pycnidium and extending toward the central cavity.

FIG. 3. Coniothyrium Spores.

As the spores accumulate pressure is exerted upon the cuticle, causing its rupture, and the spores are freed. Frequently, if the cuticle be thick, the pycnidia open under the cuticle and the spores are unable to escape until further disintegration proceeds.

The fungus is clearly a species of Coniothyrium and agrees well in all of its characters with C. *Fuckelii* Sacc. Although that species is recorded as inhabiting branches only, it is probable that it is iden-

tical with the form in question. Isolation of the fungus for the purpose of more closely studying its character was easily affected by breaking a pycnidium in sterile water and then using a cubic centimeter or so of the solution in each tube of the medium used for plating; other isolations were also made by making transfers to poured agar plates from the flesh just underneath the epidermis of the diseased spot.

In artificial culture on pea agar the fungus grows rapidly, producing a pure white mycelium. On apple agar the mycelium is tawny. Pycnidia developed upon plate cultures with extreme rarity, many cultures showing no pycnidia whatever, others a very few. In all instances where they were produced it was as the fungus became old and the agar dense or nearly dry. In some instances their arrangement upon the plate was strikingly regular and inexplicable. In one plate (Fig. 4) an almost perfect circle of pycnidia was formed at a distance of about 2.5 cm. from the point of inoculation, and

FIG. 4.—Coniothyrium Showing Arrangement of Pycnidia.

though the mycelium proceeded to grow beyond this point no further pycnidia formation occurred.

Inoculated upon sterile apple twigs the mycelium was dense and white and pycnidia were produced in abundance.

CONIOTHYRIUM ON APPLE TWIGS.

Many instances of this fungus occurring upon apple twigs have come under observation during the past year. The affected twig, usually terminal portions of recent growth, dies, the bark shrinks and shrivels, turns black and at the line of demarkation between diseased and healthy tissue cracks. (Fig. 5.) The general appearance of such a twig is much like that caused by the fire blight, but is readily distinguished by the presence of very numerous, minute, pimple-like pustules, the pycnidia, distributed over the dead part.

The significance of this fungus upon twigs is twofold: 1, as a

FIG. 5.—Coniothyrium.

cause of the twig blight, 2, as a source of infective material, spores, to transmit the rot to the fruit.

TREATMENT.

Spraying coupled with careful pruning out and burning of diseased twigs and branches will doubtless serve as a means of control of Coniothyrium both upon branches and fruit.

SPHAEROPSIS ON APPLE TWIGS.

This fungus, long known as the cause of the black rot,[1] of the fruit and later as the cause of cankers of the limbs,[2] frequently occurs

[1] Peck 34th Ann'l Rept., p. 36, 1881. [2] Paddock N. Y. Exp. Station Bulletin, 163, Dec., 1899.

To determine with certainty whether this fungus was the true cause of the scurf, also whether it could gain entrance through uninjured bark, a series of inoculations was made upon the apple trees growing at the Station farm.

Some three hundred inoculations by Mr. W. F. Kirkpatrick and Mr. S. W. Foster were made by cutting from agar plates, with a sterile knife, small squares of agar bearing the fungous threads. Inoculations were made upon bruised bark and upon unbruised bark, resulting in the production of the typical diseased condition in 49.4 per cent of the inoculations upon bruised bark and only about 3 per cent of infection upon uninjured bark. While definite proof is lacking, it is probable that all infections upon unbruised bark were through lenticels.

TREATMENT.

Spraying and close pruning of affected branches will probably lead to control of this disease.

ERRATA.—Transpose pages 53 and 55.

APPLE SCURF.[1]

BY F. L. STEVENS.

This is the name proposed for a disease which does not seem to have been heretofore described in the literature of apple diseases. The disease has been under observation for about two years in this Station, it having been sent to the Station with inquiries from two different sections of the State during that time.

The first specimen was received from Watauga County, N. C., in May, 1905. The chief symptom is a shrinking of the bark, including the epidermis, in such manner that minute air spaces are left immediately below the cuticle, giving to the affected region a silvery gray appearance. Close examination, especially with the aid of a lens, shows the presence of myriads of minute black pycnidia scattered throughout the affected region. Such a condition may extend over large areas on twigs one, two or three years of age. The seriousness of the disease, the amount of damage done, is unknown. That the condition has attracted the attention of practical apple growers to sufficient extent to cause them to submit specimens for diagnosis indicates that the injury is at least appreciable to the apple growers of the State. Death of the infected twigs does not always seem to follow infection. The disease is limited to the bark and would, therefore, be slow to cause death, and if it did it would be only through injury to the tree as a whole and would be gradual and insidious.

The black pycnidial bodies, referred to above, were found to contain minute spores, elliptical or oblong in shape, hyaline and about 2 x 5.8 Mu in size. The pycnidia were 100 to 125 Mu in diameter, black and provided with a minute ostiolum, from which the spores issue in coils upon being wet. The fungus clearly belongs either to the genus Phyllostica or Phoma, of which there are at least 22 species reported as growing upon the apple or its close kin. Since this species agrees closely with the description of P. *prunicola,* except in slight details, as regards the spot produced, and in the fact that P. *prunicola* is a leaf fungus, it seems best to regard it as being that species. Plate cultures in apple bark agar were made from spores in the spring of 1905, leading to a ready isolation of the fungus, which grew rapidly upon this medium.

It fruited in abundance, producing its characteristic pycnidia and spores. The colonies, owing to the color of the mycelium, were of striking brick-red color.

1 Attention was called to this disease at the Spring Meeting of the North Carolina Academy of Science in May, 1905, and in the Report of the Biological Division in the Annual Report of this Station for 1904-'05, pp 20, 21.

on apple twigs as a cause of blight. Several cases came under observation in this State in 1905 and were reported in the annual report of this Station. They have also been noted by Rolfs.

In general appearance Sphaeropsis on twigs is precisely like that of Coniothyrium, as described above, with the possible difference that the pycnidia are larger and less numerous. The two can not, however, be safely distinguished without microscopic examination.

TREATMENT.

The presence of Sphaeropsis as a twig blight as well as a canker causing fungus serves, as in the case with Coniothyrium, to give this fungus wider dissemination, and indicates the necessity of most careful pruning to thereby lessen the possibility of fruit infection, as well as to prevent injury from the fungus as a twig blight.